CHAPTER 13

GASTROINTESTINAL ASSESSMENT

INTRODUCTION

This system assessment centers on reported peptic ulcer and liver disease, and current hepatic function and porphyria as determined by comprehensive laboratory testing. The liver is a major target organ for single high-dose and continued low-dose exposure to chlorophenols and TCDD. Peptic/stomach ulcer disease and porphyria cutanea tarda (PCT) are suspected clinical endpoints following moderate- to high-level exposures.

A variety of experimental animal studies[1-5] have demonstrated hepatic dysfunction and porphyria following a wide range of exposures to TCDD. The effects of exposure, as measured by enzymatic change, however, generally appear to be more related to species than to dose and route of administration.

Gross organ pathology in the digestive system and associated clinical symptoms have been observed following TCDD oral administration to (or accidental ingestion by) animals. Pathological lesions have included gastric ulcers, metaplasia of the gastric mucosa, ileitus, hepatic hypertrophy and degeneration, hepatic parenchymal cell necrosis, and hepatic lipid accumulation.

Scientific interest has centered on changes in hepatic enzymes following TCDD administration. Clearly, TCDD has proved to be an exceptional inducer of hepatic enzymes and mixed function oxidases, and a powerful inhibitor of other enzymes. Specifically, the induction of cytochrome P-450, a ferrocytochrome enzyme, by TCDD has been demonstrated in many species and most of their tissues. Further, marked increases in cytochrome P-450 have been implicated in the mechanism of hepatotoxicity, although other factors, such as genetic susceptibility via the Ah locus, iron levels, and lipid peroxidation (but not vitamin A), are also contributory.[6-8]

TCDD has also been shown to produce hepatic porphyria in animals by a reduction in uroporphyrinogen decarboxylase, possibly due to the activation of the P-450 enzyme.[9,10] The porphyriogenic effect of TCDD has also been influenced by genetic susceptibility, iron levels, sex, and ambient temperature.[11,12] In correlation with some human studies, hexachlorobenzene was found to be more porphyriogenic than TCDD.[11]

Numerous morbidity studies, predominantly from the industrial sector, have noted significant abnormal liver function in exposed workers, with and without the presence of clinical hepatic disease. Abnormal liver function test results have been found for direct bilirubin, alkaline phosphatase, triglycerides, cholesterol, serum glutamic-oxaloacetic transaminase (SGOT), gamma-glutamyl transpeptidase (GGTP), urine d-glucaric acid, etc.[13-26] The consistent finding of elevated cholesterol levels may have predictive significance with respect to future heart disease (see Chapter 15), but at present there is no evidence for this.

Contemporary studies have focused on two indirect measures of hepatic microsomal activity, GGTP and urine d-glucaric acid. In the study of the English industrial incident, several Seveso investigations, and the two studies of the Monsanto plant in Nitro, West Virginia, there was modest agreement in observing elevated GGTP and urine d-glucaric acid levels in exposed individuals.[18,19,21,22] Common to all studies was the observation that individuals with chloracne manifested significantly more abnormal liver function tests than exposed individuals without chloracne or unexposed individuals, suggesting a link to TCDD exposure.

Several industrial studies have shown altered porphyrin excretion patterns (predominantly an increase in uroporphyrin) or clinical evidence of PCT, particularly in chronically exposed workers.[27-29] Individuals with low chronic exposure or high acute exposure (Seveso) have not shown these signs. Further, detailed reviews of the suspected association have identified the following scientific study design and interpretive problems: (1) multiple etiologies of PCT or abnormal porphyrin excretion patterns (chemical exposure, genetic makeup, alcohol consumption), (2) misdiagnosis of PCT, and (3) confounding of chemical exposures for the industrial cohorts.

Some investigators believe that the PCT cases found in the early U.S. and European studies were more likely caused by exposure to chlorobenzenes than to TCDD.[30] Overall, the evidence at present is inconclusive to establish a causal association between PCT and TCDD exposure.

A recent industrial study based on questionnaire data has suggested an association of stomach/peptic ulcers with exposure to TCDD.[22] This finding at the Monsanto plant differs from similar research using a slightly different cohort at the same plant which produced a negative conclusion on peptic ulcer disease.[31] The gastric ulcer-TCDD association has not been reported in other cohort dioxin morbidity studies, but ulcer disease has generally not been a major research focus. The preliminary gastric ulcer-TCDD association is fortified somewhat by studies that have shown significant gastric mucosal damage in monkeys following oral administration of TCDD.[2]

Baseline Summary Results

The 1982 AFHS examination conducted an extensive evaluation of hepatic status by questionnaire, physical examination, and laboratory testing. The questionnaire elicited data on liver conditions, liver disease, and symptoms compatible with PCT, as well as detailed information on PCT risk factors (e.g., alcohol consumption, chemical exposures). The physical examination measured hepatomegaly when present and determined liver function and porphyrin patterns by a comprehensive battery of 12 laboratory tests.

The questionnaire showed that Ranch Hands reported more miscellaneous liver conditions (verified by medical record reviews) and more skin changes compatible with PCT than their Comparisons. Although the PCT-reported data were statistically significant, no cases of PCT were diagnosed at examination in either cohort.

The physical examination detected a twofold increase in hepatomegaly in the Ranch Hands, but the numbers were small and not statistically significant. Many of the laboratory test results demonstrated statistical interactions with the covariates. These interactions can be interpreted as being

suggestive of an herbicide effect. Ranch Hands had slightly higher GGTP and lactic dehydrogenase (LDH) results and lower cholesterol levels; no differences were found for bilirubin or alkaline phosphatase levels.

SGOT, serum glutamic-pyruvic transaminase (SGPT), and LDH results in the Ranch Hands interacted with the covariates alcohol, degreasing chemicals, and industrial chemicals differently than they did in the Comparisons. All of these two-factor interactions were statistically significant ($p<0.05$). There were no significant group differences in uroporphyrin, coproporphyrin, or d-aminolevulinic acid levels, nor did any test set support a diagnosis of PCT. Exposure analyses were essentially negative.

The comprehensive hepatic evaluation did not reveal any consistent pattern of significant liver damage in the Ranch Hand group. Nevertheless, because of subtle profile differences in conjunction with questionnaire results and recent literature citations, the gastrointestinal system continues to be targeted for intensive examination throughout all phases of the followup effort.

Parameters of the 1985 Gastrointestinal Assessment

The 1985 AFHS examination continued the emphasis on hepatic function and expanded the porphyrin test battery to six assays. In addition, new components were added to the questionnaire to assess past and current diagnosed peptic ulcer disease, along with a series of screening questions to assess possible undiagnosed disease. Covariate data on aspirin usage, blood group, and family history of peptic ulcer were likewise obtained. Additional probes on intestinal parasites, gallbladder disease, and other liver conditions were also added. Because of the known profound effects of alcohol ingestion on hepatic function, a detailed alcohol consumption history was obtained by questionnaire.

Thus, the dependent variables and covariates in the analyses below reflect a substantial enhancement over those assessed in the 1982 Baseline examination. Because of the effects of increased body temperature and past/current hepatitis B on some liver function tests, participants with a fever of 100 or more degrees Fahrenheit and/or a positive hepatitis B surface antigen (HB_sAg) test were excluded from the analyses. Categorization of continuous clinical variables to dichotomous variables was largely accomplished by use of normal test values from the SCRF laboratory. Minor numeric differences in the tables that follow are due to an occasionally missing value.

The analyses are generally based on 1,009 Ranch Hands and 1,289 total Comparisons after removal of the febrile and positive HB_sAg participants. The statistical analyses relied largely on general linear models (SAS®-GLM), logistic regression techniques (BMDP®-LR), and log-linear models (BMDP®-4F). Parallel analyses using Original Comparisons are found in Tables K-7 to K-16 of Appendix K.

RESULTS AND DISCUSSION

This chapter, entitled "Evaluation of Hepatic Status" in the Baseline Report, incorporates the new elements of peptic ulcer disease and mortality from diseases of the digestive system; hence, the chapter name change to "Gastrointestinal Assessment."

Because of the importance of gastrointestinal disorders, numerous historical and laboratory variables were chosen for evaluation. The analyses are reported in the following order: questionnaire data, mortality data, physical examination findings, laboratory results, exposure index analyses, and representative longitudinal analyses.

Questionnaire Data: Liver Disorders

At the followup examination, each participant was asked whether he had developed hepatitis, jaundice, cirrhosis, or other liver disorders during the interval 1982 to 1985. Affirmative responses were subsequently subject to verification by medical record reviews.

Since the Baseline interview, eight Ranch Hands and five Comparisons cited a verified history of hepatitis (p=0.264); four Ranch Hands and five Comparisons reported a subsequently verified history of enlarged liver (p=0.999); one from each group noted a verified symptom of jaundice; one Ranch Hand cited a confirmed interval history of cirrhosis; and six Ranch Hands and six Comparisons gave verified histories of seven miscellaneous liver disorders (p=0.774). Table 13-1 presents the ICD code and descriptive diagnosis of the miscellaneous liver disorders by group.

Because the number of respondents with new liver disorders was small and precluded meaningful analyses, the verified interval history was added to the verified Baseline history to assess possible lifetime differences for liver disease. These combined results are presented in Table 13-2.

On the basis of combined data, the verified questionnaire responses for historic hepatitis, jaundice, cirrhosis, enlarged liver, and miscellaneous liver disorders did not vary significantly between the Ranch Hand and Comparison groups. The results for miscellaneous liver disorders differed from the Baseline findings. At Baseline, significantly more Ranch Hands than Original Comparisons had a verified liver disorder other than jaundice, hepatitis, or cirrhosis (13/1,045 versus 1/773; p=0.006). Subsequent to Baseline, the status of one additional Ranch Hand disorder and one more Original Comparison disorder was verified. Including these two new verified conditions with the data from replacement and shifted Comparisons, the group contrast at Baseline would have been of borderline significance (14/1,045 versus 7/1,224; p=0.077). Combining these Baseline data with the followup data resulted in nonsignificant lifetime results. However, the combined Baseline and interval analysis contrasting the Ranch Hands and the Original Comparisons was marginally significant (p=0.065) due to the contribution of the significant Baseline results.

The verification status of reported liver symptoms and diseases is presented in Table 13-3. The data reflect the proportions of historic reporting that were verified by medical record reviews, and are contrasted by group for each variable. These data showed that the proportion of verified disease was not statistically significant between groups except for the category of enlarged liver which showed a higher confirmation rate in the Comparison group. Thus, over-reporting or symptom/disease misclassification by the participants was not a function of group membership.

TABLE 13-1.

Number of Other Liver Conditions Reported by Study Participants at Followup by Group (Verified by Medical Record Review)

ICD* Code (Meaning)	Group	
	Ranch Hand	Comparison
5713 (Alcoholic Liver Damage)	1	1
57420 (Calculus of Gallbladder without Mention of Cholecystitis)	0	1
7891 (Hepatomegaly)	1	1
7904/7905 (Enzyme Elevation)	3	0
7948 (Abnormal Liver Scan)	1	1
E9426 (Adverse Effect of Drug)	0	1
M81406 (Adenocarcinoma)	0	1
Total	6	6

*ICD = International Classification of Diseases.

TABLE 13-2.

Unadjusted Analyses for Baseline and Interval History of Liver Disease by Group (Verified by Medical Record Review)

Disease	Statistic	Group: Ranch Hand Number	Ranch Hand Percent	Comparison Number	Comparison Percent	Est. Relative Risk (95% C.I.)	p-Value
Hepatitis (Viral and Alcoholic)	n	1,016		1,293			
	Yes	37	3.6	43	3.3	1.10 (0.70,1.72)	0.731
	No	979	96.4	1,250	96.7		
Jaundice	n	1,016		1,293			
	Yes	20	2.0	28	2.2	0.91 (0.51,1.62)	0.771
	No	996	98.0	1,265	97.8		
Cirrhosis	n	1,016		1,293			
	Yes	3	0.5	2	0.2	1.91 (0.32,11.46)	0.660
	No	1,013	99.5	1,291	99.8		
Enlarged Liver	n	1,016		1,293			
	Yes	17	1.7	24	1.9	0.90 (0.48,1.68)	0.874
	No	999	98.3	1,269	98.1		
Miscel-laneous Liver Disorders	n	1,016		1,293			
	Yes	17	1.7	13	1.0	1.68 (0.81,3.47)	0.195
	No	999	98.3	1,280	99.0		

TABLE 13-3.

Medical Record Verification of Reported Liver Symptoms and Diseases by Group (Baseline and Interval Questionnaires Combined)

Variable	Verification Status	Group		p-Value
		Ranch Hand	Comparison	
Hepatitis	Number Reported	47	53	
	Medical Records Reviewed	44	48	
	Medical Records Pending or Not Released	3	5	
	Number Verified	37	43	
	Percent Verified	78.7	81.1	0.806
Jaundice	Number Reported	43	59	
	Medical Records Reviewed	23	35	
	Medical Records Pending or Not Released	20	24	
	Number Verified	20	28	
	Percent Verified	46.5	47.5	0.999
Cirrhosis	Number Reported	7	3	
	Medical Records Reviewed	5	3	
	Medical Records Pending or Not Released	2	0	
	Number Verified	3	2	
	Percent Verified	42.9	66.7	0.999
Enlarged Liver	Number Reported	30	29	
	Medical Records Reviewed	29	29	
	Medical Records Pending or Not Released	1	0	
	Number Verified	17	24	
	Percent Verified	56.7	82.8	0.047
Miscellaneous Liver Disorders	Number Reported	21	14	
	Medical Records Reviewed	20	14	
	Medical Records Pending or Not Released	1	0	
	Number Verified	17	13	
	Percent Verified	94.4	92.9	0.627

Peptic Ulcer Diseases

The primary purpose of these analyses was to compare the ulcer disease experience of the Ranch Hand and Comparison groups. Since blood type has been reported to affect the incidence of peptic ulcer disease, blood type was used as a covariate in these analyses. The military medical and personnel records of the 2,309 study participants were reviewed to determine the blood type as recorded in these sources. The distribution of blood types in the two groups is shown in Table 13-4.

TABLE 13-4.

Unadjusted Analysis of Blood Type by Group

	Blood Type								
	O		A		B		AB		
Group	Number	Percent	Number	Percent	Number	Percent	Number	Percent	Total*
Ranch Hand	378	45.4	334	40.1	87	10.5	33	4.0	832
Comparison	504	46.4	425	39.1	125	11.5	33	3.0	1,087
				p=0.60					

*184 Ranch Hands and 206 Comparisons missing from blood type analysis.

The blood type distribution was not significantly different in the two groups (p=0.60), and was similar to the distribution of blood types in the general U.S. white male population (p=0.57).

Both physical examination diagnoses and questionnare responses to questions concerning ulcers were used as sources of data on the occurrence of ulcer disease. A total of 58 participants was diagnosed as having ulcer disease at the time of the examination; however, 13 had to be deleted from the analyses of physical examination data and 15 from the analyses of questionnaires due to missing data on blood type. On questionnaires, 42 reported currently having ulcers and an additional 126 reported having had ulcers in the past. These data are summarized in Table 13-5.

A three-factor log-linear analysis (group, ulcer, blood type) of data from the physical examination showed a significant three-factor interaction, with the Ranch Hand rate being higher in blood types AB and O, and lower for types A and B (p=0.03). Stratified analyses of each blood type were conducted and did not reveal any statistically significant group differences. These data are shown in Table 13-6.

TABLE 13-5.

Frequency of Diagnosed and Reported Ulcer Disease by Group

		Group				
		Ranch Hand		Comparison		
Variable	Statistic	Number	Percent	Number	Percent	Total
Diagnosed Disease	n	832		1,087		1,919
(Physical	Yes	19	2.3	26	2.4	45
Examination Data)	No	813	97.7	1,061	97.6	1,874
Reported Disease	n	832		1,085		1,917
(Questionnaire	Current	22	2.6	20	1.8	44
Data)	Past	53	6.4	73	6.7	126
	None	757	91.0	992	91.4	1,749

A three-factor log-linear analysis of questionnaire data was also performed. This analysis looked at current and past history of ulcer disease. No significant group differences or multifactor interactions were seen, with all p-values being greater than 0.10.

These analyses demonstrated overall group equivalence within the Ranch Hand and Comparison groups with respect to blood type and present and past ulcer disease.

Mortality Count Data

Linkage of digestive system mortality to observed historic or examination morbidity has not been explored in this report; the linkage process, with the use of the Comparison replacement strategy, remains an open research issue. From a broader perspective, however, review of mortality count data in conjunction with current morbidity data may be useful in identifying disease pattern(s) with respect to group membership, organ-specific disease, and important covariates. For these purposes, the latest mortality count data (as of 31 December 1985) are summarized in Table 13-7.

These data showed a large mortality contribution (approximately 50%)from liver disease in both groups and a relative excess in Ranch Hands as contrasted to Comparisons. For malignant neoplasms, there was a relative excess in the Comparison group. There is also the suggestion that alcohol is an important risk factor. The relative excess of malignant neoplasms in the Comparison group is also striking. Overall, the slight excess of digestive system mortality in the Ranch Hands and the differences in distribution of

TABLE 13-6.

Unadjusted Analyses of Peptic Ulcer Disease by Blood Type by Group

		Group					
		Ranch Hand		Comparison			
Blood Type	Statistic	Number	Percent	Number	Percent	Est. Relative Risk (95% C.I.)	p-Value
O	n	378		504			
	Yes	13	3.4	11	2.2	1.60 (0.70,3.60)	0.37
	No	365	96.6	493	97.8		
A	n	334		425			
	Yes	4	1.2	12	2.8	0.42 (0.14,1.29)	0.21
	No	330	98.8	413	97.2		
B	n	87		125			
	Yes	0	0.0	3	2.4	--	0.27[a]
	No	87	100.0	122	97.6		
AB	n	33		33			
	Yes	2	6.1	0	0.0	--	0.49[a]
	No	31	93.9	33	100.0		

--Estimated relative risk and confidence interval not calculated due to zero count in a cell.

[a]Fisher's exact test.

TABLE 13-7.

Frequency of Digestive System Mortality by Group

ICD Code	Deaths, by Group	
	Ranch Hand	1:5 Comparison
Pancreatitis (5770)	1	2
Alcoholic cirrhosis (5712)	0	6
Nonalcoholic cirrhosis (5715)	3	5
Nonalcoholic fatty liver (5718)	0	1
Chronic liver disease (5728)	1	1
Alcoholic liver disease (5711)	1	0
Duodenal ulcer (5325)	0	1
Malignant neoplasm (150-159)	2	15
Total	8	31

deaths by cause in the two groups raise the issue of competing mortality. Interpretation of the analyses in this report of hepatic function and liver disease, with alcohol consumption taken into account, should be reviewed in the light of these mortality data.

Physical Examination Data

Gastrointestinal dysfunction was not a major focus of the physical examination except for a comprehensive biochemical profile of the liver. Consequently, only data on hepatomegaly were analyzed, and results of the analysis are shown in Table 13-8.

The analysis showed a marginally significant excess (eight cases versus three) of hepatomegaly in the Ranch Hands ($p=0.069$). These results were in relative contrast to the Baseline examination findings of 1.56 percent and 0.78 percent in the Ranch Hand and Comparison groups, respectively ($p=0.138$), in the sense that fewer abnormalities were detected at the followup, although at both examinations the difference favored the Comparisons.

The group data for hepatomegaly were pooled and compared to the covariates of age, race, occupation, current alcohol use (one or less drinks per day, more than one to four drinks per day, and more than four drinks per day), lifetime exposure to industrial chemicals, and lifetime exposure to degreasing chemicals. Only age and occupation showed significant associations with hepatomegaly ($p=0.018$, $p=0.026$, respectively). Because of sparse data, an adjusted analysis was not conducted.

General Laboratory Examination Data

As in the Baseline Report, the followup examination emphasized evaluation of laboratory data, particularly for hepatic function. Thus, this

TABLE 13-8.

Unadjusted Analysis of Enlarged Livers Diagnosed at Physical Examination by Group*

	Enlarged Liver					
	Yes		No			
Group	Number	Percent	Number	Percent	Total	p-Value
Ranch Hand	8	0.8	1,002	99.2	1,010	0.069
Comparison	3	0.2	1,287	99.8	1,290	

*Excludes participants with positive HB_sAg.

section reports on nine laboratory tests of hepatic function and on two tests reflecting porphyrin metabolism. Normal ranges for these 11 variables as determined by the SCRF and the Mayo Clinic Laboratories are presented in Table 13-9. Only values greater than the normal range were considered important in the assessment of dysfunction.

Analyses of the nine hepatic variables were adjusted for the covariates of age, race, occupation (OCC), current alcohol use (ALC), days of exposure to industrial chemicals (IC), and days of exposure to degreasing chemicals (DC). For the two porphyrin analyses, blood urea nitrogen was used as a covariate. Because the hepatic test variables encompass acute to chronic effects, there was no "ideal" alcohol covariate (e.g., drink-years, current alcohol consumption in drinks per day).

The covariate alcohol use was obtained from questionnaire data, centering on daily alcohol consumption (beer, wine, liquor) for those participants who reported drinking at least one drink in the 2 weeks preceding the examination. Thus, the alcohol covariate measures recent drinking intensity and may be more useful in adjustment of acute variables (e.g., GGTP, SGPT) than variables related to chronic liver dysfunction (e.g., bilirubin determinations, alkaline phosphatase).

Exposure to industrial chemicals and degreasing chemicals was measured in cumulative days of unprotected exposure, and was derived from the 1982 and 1985 questionnaires. These data, therefore, represent lifetime exposure.

Exclusion categories consisted of fever (over 100 degrees Fahrenheit) and positive HB_sAg tests, because of the known effects of these conditions on liver function tests. Three participants (two Ranch Hands, one Comparison) were excluded because of fever, and eight (five Ranch Hands, three Comparisons) because of a positive HB_sAg test (seven positive, one missing). In addition, due to missing alcohol data, nine other individuals (six Ranch Hands, three Comparisons) were deleted from the analyses when current alcohol use was found to be a significant covariate.

TABLE 13-9.

Laboratory Norms for Nine Hepatic Function Variables and Two Porphyrin Determinations

Variable	Unit	SCRF Normal	SCRF Abnormal
SGOT	U/L	27-47	≥48
SGPT	U/L	3-36	≥37
GGTP	U/L	15-85	≥86
Alkaline Phosphatase	U/L	50-136	≥137
Total Bilirubin	mg/dl	≤1.5	>1.5
Direct Bilirubin	mg/dl	≤0.36	≥0.37
LDH	U/L	100-190	≥191
Cholesterol[a]	mg/dl	≤260	≥261
Triglycerides[a]	mg/dl	≤320	≥321
Uroporphyrin[b]	mg/24 hrs	≤46	≥47
Coproporphyrin[b]	mg/24 hrs	≤96	≥97

[a]SCRF provides age-dependent normal ranges; these values represent the maximum normal limits for those older than 40.

[b]Performed at the Mayo Clinic.

Statistical Analyses

The nine dependent variables from the hepatic battery were subjected to three types of basic analyses: (1) a continuous dependent variable adjusted by continuous covariates (CC), (2) a continuous dependent variable adjusted by discrete covariates (CD), and (3) a discrete (categorical) dependent variable adjusted by discrete covariates (DD), except for current alcohol use, which was left as a continuous variable for model-fitting and power purposes. General linear models (SAS®) were used for the CC and CD analyses, and BMDP®-LR was used for the DD analyses.

As noted in Chapter 7, Statistical Methods, all adjustments were carried out with the simplest model, including all significant covariates and two- and three-way interactions. The log transformation was used for the nine hepatic variables and for uroporphyrin, while a square root transformation was employed for the coproporphyrin variable. Since some direct bilirubin values were 0, the value 0.10 was added prior to log transformation.

The sample sizes were sufficient to detect a 1.93-fold increase in the frequency of abnormal values for alkaline phosphatase and a 1.42-fold increase in the frequency of abnormal values for SGPT, using a (two-sided) α-level of 0.05 and power 0.80. Further, the sample sizes were sufficient to detect a 0.7 percent mean shift in alkaline phosphatase, a 1.8 percent mean shift in SGPT, and a 2.8 percent mean shift in uroporphyrin values.

The results of the analyses on the 11 dependent variables are presented in the following summary tables (Tables 13-10 through 13-12), followed by descriptive narrative text. The summary tables are in the following logical order: unadjusted results, covariate tests of association, and adjusted results. Tables K-1 and K-2 of Appendix K summarize interactions from the statistical analyses. All analytic information on any given variable can be obtained by scanning the summary tables.

The following discussion condenses the key information on each dependent variable. Group-by-covariate interactions are narratively presented. The variables are organized in the same order as given in the tables.

Serum Glutamic-Oxaloacetic Transaminase (SGOT)

The unadjusted continuous (group means) and categorical (percent abnormalities) tests showed no statistically significant differences between groups ($p=0.298$ and $p=0.999$, respectively).

Tests of association with the covariates using pooled group categorical data demonstrated the significant effect of race (a higher percentage of abnormalities in Blacks than nonblacks, 13.5% versus 7.6%; $p<0.022$) and current alcohol use (21.2% abnormal values associated with more than four drinks per day, 9.0% abnormals for more than one to four drinks per day, and 5.8% for one or less drinks per day; $p<0.001$). Similarly, the mean SGOT levels differed significantly between races ($p<0.001$) and by current alcohol use ($p<0.001$).

The CC adjusted model showed no significant group differences ($p=0.309$). Significant covariates were race, an interaction of current alcohol use-by-degreasing chemicals, and an interaction of current alcohol use-by-age (all

TABLE 13-10.

Unadjusted Continuous and Categorical Analyses for Hepatic Function Variables and Two Porphyrin Determinations by Group

Variable	Statistic	Group: Ranch Hand	Group: Comparison	Est. Relative Risk (95% C.I.)	p-Value
SGOT	n	1,009	1,289		
	Mean	33.5	33.0		0.298
	95% C.I.	(32.8,34.1)	(32.5,33.5)		
	Number/%				
	Normal	929 92.1%	1,187 92.1%	1.00 (0.74,1.36)	0.999
	High	80 7.9%	102 7.9%		
SGPT	n	1,009	1,289		
	Mean	21.6	22.5		0.051
	95% C.I.	(20.9,22.3)	(21.9,23.1)		
	Number/%				
	Normal	872 86.4%	1,102 85.5%	0.93 (0.73,1.17)	0.546
	High	137 13.6%	187 14.5%		
GGTP	n	1,009	1,289		
	Mean	32.8	32.4		0.632
	95% C.I.	(31.4,34.3)	(31.2,33.6)		
	Number/%				
	Normal	919 91.1%	1,172 90.9%	0.98 (0.74,1.31)	0.942
	High	90 8.9%	117 9.1%		
Alkaline Phospha-tase	n	1,009	1,289		
	Mean	91.8	89.3		0.009
	95% C.I.	(90.4,93.3)	(88.1,90.6)		
	Number/%				
	Normal	953 94.5%	1,236 95.9%	1.37 (0.93,2.01)	0.114
	High	56 5.6%	53 4.1%		
Total Bilirubin	n	1,009	1,289		
	Mean	0.74	0.75		0.576
	95% C.I.	(0.73,0.76)	(0.74,0.76)		
	Number/%				
	Normal	982 97.3%	1,250 97.0%	0.88 (0.54,1.45)	0.706
	High	27 2.7%	39 3.0%		

TABLE 13-10. (continued)

Unadjusted Continuous and Categorical Analyses for Hepatic Function Variables and Two Porphyrin Determinations by Group

Variable	Statistic	Group: Ranch Hand	Group: Comparison	Est. Relative Risk (95% C.I.)	p-Value
Direct Bilirubin	n	1,009	1,289		
	Mean	0.18	0.18		0.981
	95% C.I.	(0.17,0.18)	(0.17,0.18)		
	Number/%				
	Normal	971 96.2%	1,246 96.7%	1.13 (0.73,1.77)	0.649
	High	38 3.8%	43 3.3%		
LDH	n	1,009	1,289		
	Mean	123.5	123.9		0.655
	95% C.I.	(122.2,124.8)	(122.7,125.2)		
	Number/%				
	Normal	999 99.0%	1,272 98.7%	0.75 (0.34,1.64)	0.560
	High	10 1.0%	17 1.3%		
Cholesterol	n	1,009	1,289		
	Mean	214.3	215.0		0.688
	95% C.I.	(211.8,216.8)	(212.8,217.2)		
	Number/%				
	Normal	863 85.5%	1,082 83.9%	0.88 (0.70,1.11)	0.322
	High	146 14.5%	207 16.1%		
Triglycerides	n	1,009	1,289		
	Mean	118.5	117.3		0.719
	95% C.I.	(113.8,123.3)	(113.4,121.4)		
	Number/%				
	Normal	941 93.3%	1,210 93.9%	1.11 (0.79,1.55)	0.549
	High	68 6.7%	79 6.1%		
Uroporphyrin	n	1,006	1,286		
	Mean	16.9	17.9		0.048
	95% C.I.	(16.2,17.7)	(17.3,18.6)		
Copropor-phyrin	n	1,008	1,287		
	Mean	119.1	115.6		0.081
	95% C.I.	(116.2,122.0)	(113.0,118.2)		

TABLE 13-11.

Association Between Nine Hepatic Function Variables and Two Porphyrin Determinations and Six Covariates in the Combined Ranch Hand and Comparison Groups

Variable	Analysis*	Age	Race	Occupation	Alcohol	Industrial Chemicals	Degreasing Chemicals
SGOT	C	NS	<0.001	NS	<0.001	NS	NS
	D	NS	0.022	NS	<0.001	NS	NS
SGPT	C	<0.001	NS	NS	<0.001	NS	0.017
	D	0.001	NS	NS	<0.001	NS	NS
GGTP	C	0.012	<0.001	0.032	<0.001	NS	NS
	D	NS	0.021	NS	<0.001	NS	NS
Alkaline Phosphatase	C	NS	NS	<0.001	<0.001[a]	<0.001	0.010
	D	NS	NS	0.003	NS*[a]	0.030	NS*
Total Bilirubin	C	NS*	NS	0.011	0.008	NS	NS
	D	NS	<0.001	NS	NS	NS	NS
Direct Bilirubin	C	NS	NS*	NS	<0.001	NS	NS
	D	NS	0.015	NS	NS	NS	NS
LDH	C	<0.001	0.006	NS	NS	NS	NS
	D	NS	NS	NS	NS	NS	NS
Cholesterol	C	<0.001	NS	0.002	<0.001	NS	NS
	D	0.010	NS	0.008	0.018	NS	NS
Triglycerides	C	<0.001	<0.001	0.013	0.030	NS*	0.019
	D	NS	0.031	NS	NS	NS*	NS
Uroporphyrins	C	NS	NS	NS	NS	NS	NS
Coproporphyrins	C	0.003	NS	NS	<0.001	NS	NS

*Continuous (C)/Discrete (D).

NS: Not significant ($p>0.10$)

NS*: Borderline significant ($0.05<p\leq 0.10$).

[a]Wine consumption.

TABLE 13-12.

Adjusted Continuous and Categorical Analyses for Hepatic Function Variables and Two Porphyrin Determinations by Group

Variable	Analysis	Statistic	Group: Ranch Hand	Group: Comparison	Adj. Relative Risk (95% C.I.)	p-Value	Covariate Remarks*
SGOT	CC	n	1,003	1,286			ALC*DC(p<0.001)
		Adj. Mean	34.8	34.3	—	0.309	AGE*ALC(p<0.001)
		95% C.I.	(33.8,35.7)	(33.4,35.3)			RACE(p<0.001)
	CD	n	1,003	1,286			GRP*ALC(p=0.048)
		Adj. Mean	****	****	—	****	ALC*IC(p=0.008)
		95% C.I.	****	****			DC(p=0.019), RACE(p<0.001)
	DD	n	1,003	1,286	1.03 (0.75,1.41)	0.868	AGE*ALC(p<0.001)
							OCC*ALC(p<0.001)
							RACE (p=0.026)
SGPT	CC	n	1,003	1,286			ALC*DC(p=0.008), RACE*DC(p=0.015)
		Adj. Mean	21.4	22.2	—	0.048	AGE*ALC(p=0.001), RACE*IC(p=0.017)
		95% C.I.	(20.4,22.4)	(21.3, 23.3)			
	CD	n	1,003	1,286			ALC*DC(p=0.032), AGE*ALC(p=0.022)
		Adj. Mean	21.9	22.9	—	0.029	OCC*AGE(p=0.026), IC(p=0.049)
		95% C.I.	(20.2,23.8)	(21.1,24.8)			
	DD	n	1,003	1,286	0.93 (0.73,1.18)	0.531	AGE*ALC(p=0.004)
GGTP	CC	n	1,003	1,286			AGE*ALC(p<0.001),RACE*IC(p=0.011)
		Adj. Mean	37.5	37.0	—	0.575	ALC*DC(p<0.001), AGE*DC(p=0.009)
		95% C.I.	(35.2,40.1)	(34.7,39.3)			
	CD	n	1,003	1,286			AGE*ALC(p=0.023), OCC*ALC(p=0.044)
		Adj. Mean	44.1	43.6	—	0.668	RACE(p<0.001)
		95% C.I.	(40.0,48.6)	(39.6,47.9)			
	DD	n	1,003	1,286	1.00 (0.74,1.34)	0.971	AGE*ALC(p<0.001), RACE(p=0.016)

TABLE 13-12. (continued)

Adjusted Continuous and Categorical Analyses for Hepatic Function Variables and Two Porphyrin Determinations by Group

Variable	Analysis	Statistic	Group: Ranch Hand	Group: Comparison	Adj. Relative Risk (95% C.I.)	p-Value	Covariate Remarks*
	CC	n	1,003	1,285			AGE*IC(p=0.010), RACE*IC(p=0.007)
		Adj. Mean	91.6	89.1	—	0.008	OCC(p<0.001), WINE(p<0.001)
		95% C.I.	(89.4,93.9)	(87.0,91.2)			
Alkaline	CD	n	1,003	1,285			GRP*IC(p=0.011), AGE*IC(p=0.019)
Phosphatase		Adj. Mean	****	****	—	****	RACE*IC(p=0.002), OCC(p<0.001)
		95% C.I.	****	****			WINE (p<0.001)
	DD	n	1,003	1,285	1.44 (0.97,2.13)	0.070	WINE*DC(p=0.006), AGE*IC(p=0.005)
							RACE*IC(p=0.004), OCC*IC(p=0.016)
	CC	n	1,003	1,286			AGE*DC(p=0.039)
		Adj. Mean	0.78	0.78	—	0.599	RACE*ALC(p=0.007)
		95% C.I.	(0.75,0.81)	(0.75,0.81)			RACE*OCC(p=0.001)
Total	CD	n	1,003	1,286			RACE*ALC(p=0.004)
Bilirubin		Adj. Mean	0.83	0.83	—	0.598	OCC*ALC(p=0.034)
		95% C.I.	(0.79,0.87)	(0.80,0.87)			OCC*RACE(p=0.002)
	DD	n	1,009	1,289	0.89 (0.54,1.47)	0.648	RACE(p<0.001)
	CC	n	1,003	1,286			
		Adj. Mean	0.18	0.18	—	0.972	RACE*ALC(p=0.025)
		95% C.I.	(0.17,0.20)	(0.17,0.19)			
Direct	CD	n	1,003	1,286			DC*IC(p=0.025), ALC*DC(p=0.012)
Bilirubin		Adj. Mean	0.21	0.20	—	0.830	RACE*ALC(p=0.019), OCC*ALC(p=0.002)
		95% C.I.	(0.19,0.22)	(0.19,0.22)			
	DD	n	1,003	1,286	****	****	GRP*IC(p=0.012), RACE(p=0.014)
							ALC(p=0.026)

TABLE 13-12. (continued)

Adjusted Continuous and Categorical Analyses for Hepatic Function Variables and Two Porphyrin Determinations by Group

Variable	Analysis	Statistic	Group: Ranch Hand	Group: Comparison	Adj. Realtive Risk (95% C.I.)	p-Value	Covariate Remarks*
LDH	CC	n	1,003	1,286			GRP*AGE(p=0.018), OCC*IC(p=0.014)
		Adj. Mean	****	****	—	****	RACE*IC(p=0.024)
		95% C.I.	****	****			
	CD	n	1,003	1,286			RACE(p<0.001), AGE(p<0.001)
		Adj. Mean	130.0	130.5	—	0.671	DC(p=0.016)
		95% C.I.	(127.3,132.8)	(127.8,133.1)			
	DD	n	1,009	1,289	0.75 (0.34,1.64)	0.560	
Cholesterol	CC	n	1,003	1,286			RACE*DC(p=0.021), RACE*OCC(p=0.005)
		Adj. Mean	219.5	220.4	—	0.604	IC(p=0.043), ALC(p<0.001)
		95% C.I.	(214.5,224.7)	(215.5,225.4)			AGE(p<0.001)
	CD	n	1,003	1,286			RACE*OCC(p=0.027), ALC(p<0.001)
		Adj. Mean	223.8	224.9	—	0.548	AGE(p<0.001)
		95% C.I.	(217.7,230.1)	(218.9,231.0)			
	DD	n	1,003	1,286	0.85 (0.68,1.08)	0.181	RACE*ALC(p=0.012), AGE(p=0.029)
							OCC(p=0.039)
Triglycerides	CC	n	1,003	1,286			GRP*AGE(p=0.015), ALC*DC(p=0.005)
		Adj. Mean	****	****	—	****	RACE*ALC(p=0.031), OCC(p<0.001)
		95% C.I.	****	****			
	CD	n	1,003	1,286			OCC(p<0.001), RACE(p<0.001)
		Adj. Mean	112.5	112.1	—	0.905	AGE(p<0.001), ALC(p=0.038)
		95% C.I.	(103.7,121.9)	(103.7,121.2)			
	DD	n	1,009	1,289	****	****	GRP*OCC(p=0.027), RACE (p=0.026)
							IC(p=0.038)

TABLE 13-12. (continued)

Adjusted Continuous and Categorical Analyses for Hepatic Function Variables and Two Porphyrin Determinations by Group

Variable	Analysis	Statistic	Group: Ranch Hand	Group: Comparison	Adj. Relative Risk (95% C.I.)	p-Value	Covariate Remarks*
Uroporphyrin	CC	n	1,000	1,283			GRP*BUN(p=0.015)
		Adj. Mean	****	****	—	****	DC*OCC(p=0.005)
		95% C.I.	****	****			ALC(p=0.026)
Coproporphyrin	CC	n	1,002	1,284			AGE*ALC(p=0.003)
		Adj. Mean	119.3	115.7	—	0.065	BUN(p<0.001)
		95% C.I.	(116.4,122.2)	(113.2,118.2)			

*Abbreviations:
GRP: group
OCC: occupation
ALC: current alcohol use
WINE: wine consumption
DC: exposure to degreasing chemicals
IC: exposure to industrial chemicals
BUN: blood urea nitrogen

— No relative risk or confidence interval given for continuous analyses.
**** Group-by-covariate interaction—adjusted mean/relative risk, confidence interval, and p-value are not presented.

with p<0.001). The CD analysis revealed a significant group (GRP)-by-current alcohol use interaction (p=0.048), precluding a direct group contrast. Exploration of the interaction disclosed that the Ranch Hands had a significantly higher (p=0.010) mean SGOT for the more than one to four drinks per day category, whereas there were no significant group differences for the one or less drinks per day or more than four drinks per day categories (see Table K-1 of Appendix K). Other significant covariate effects included degreasing chemicals (p=0.019), race (p<0.001), and a current alcohol use-by-industrial chemical (IC) interaction (p=0.008). The DD SGOT analysis showed no significant group differences (p=0.868). Covariates making significant contributions were race (p=0.026), an age-by-current alcohol use interaction (p<0.001), and an occupation (OCC)-by-current alcohol use interaction (p<0.001).

Serum Glutamic-Pyruvic Transaminase (SGPT)

The unadjusted categorical analysis was not significant (p=0.546), but the comparison of group means showed a borderline significant result, with the Comparisons having a higher mean SGPT than the Ranch Hands (p=0.051).

Covariate associations with the pooled categorical Ranch Hand and Comparison group data showed an inverse relationship (p=0.001) between SGPT levels and age, with 17.1 percent abnormalities for those born in or after 1942, 12.3 percent for those born between 1923 and 1941, and 8.1 percent for those born in or before 1922. The relationship with current alcohol use was also profound (p<0.001), with 23.4 percent abnormals noted for more than four drinks per day, 15.3 percent abnormals for more than one to four drinks per day, and 12.4 percent for one or less drinks per day. The direction and magnitude of the covariate effects of age and alcohol were quite similar for the tests of association with the mean SGPT level of both groups (p<0.001 for both covariates).

No significant group interactions were detected in either the discrete or the continuous analyses. The CC-adjusted analysis yielded a significant group difference, with the Comparisons having a higher group mean than the Ranch Hands (p=0.048). The model was adjusted by the interactions of current alcohol use-by-degreasing chemicals (p=0.008), current alcohol use-by-age (p=0.001), race-by-degreasing chemicals (p=0.015), and race-by-industrial chemicals (p=0.017). The CD model also showed a significantly elevated mean SGPT in the Comparison group (p=0.029). The analysis was adjusted for exposure to industrial chemicals (p=0.049), and the interactions of age-by-occupation (p=0.026), age-by-current alcohol use (p=0.022), and current alcohol use-by-degreasing chemicals (p=0.032). A borderline significant interaction (p=0.0505) between group and current alcohol use was found, but because of modeling strategy, this interaction was not included in the final model. (This interaction is explored further in Table K-1 in Appendix K, however.) The DD-adjusted analysis, like the unadjusted discrete analysis, disclosed a nonsignificant group difference (p=0.531). The model was adjusted for an age-by-current alcohol use interaction (p=0.004).

Gamma-Glutamyl Transpeptidase (GGTP)

The unadjusted contrasts of both mean levels of GGTP and the frequency of abnormalities showed no significant differences between the Ranch Hand and Comparison groups (p=0.632 and p=0.942, respectively).

For discrete covariate associations, significance was noted for race, with 14.9 percent abnormals in Blacks and 8.6 percent for nonblacks ($p=0.021$), and current alcohol use, with 26.1 percent abnormals for more than four drinks per day, 10.5 percent for more than one to four drinks per day, and 6.2 percent for one or less drinks per day use ($p<0.001$). While the mean level of GGTP was similarly affected by race and current alcohol ($p<0.001$ for both covariates), it was also influenced by age (30.3 U/L for those born in or before 1922, 33.9 U/L for those born between 1923 and 1941, and 31.1 U/L for those born in or after 1942; $p=0.012$) and occupation (31.5 U/L for officers, 35.2 U/L for enlisted flyers, and 32.5 U/L for enlisted groundcrew; $p=0.032$).

Each of the three adjusted analyses consistently produced nonsignificant group differences (CC: $p=0.575$; CD: $p=0.668$; DD: $p=0.971$). None of the three models was affected by a group-by-covariate interaction. The CC analysis was adjusted by four covariate interactions: age-by-current alcohol use ($p<0.001$), race-by-industrial chemicals ($p=0.011$), current alcohol use-by-degreasing chemicals ($p<0.001$), and age-by-degreasing chemicals ($p=0.009$). The CD model was adjusted by race ($p<0.001$), by an age-by-current alcohol use interaction ($p=0.023$), and by an occupation-by-current alcohol use interaction ($p=0.044$). The DD analysis was adjusted by race ($p=0.016$) and by an age-by-current alcohol use interaction ($p<0.001$).

Alkaline Phosphatase

The analysis of group mean values showed a significantly higher ($p=0.009$) Ranch Hand mean (91.8 U/L) than that observed in the Comparison group (89.3 U/L). The unadjusted categorical analysis revealed a higher percentage of Ranch Hand abnormalities (5.6%) than Comparison abnormalities (4.1%), but this difference was not significant (Est. RR=1.37, 95% C.I.: [0.93,2.01], $p=0.114$).

With pooled group data, significant covariate associations were found between the proportion of abnormal values and occupation ($p=0.003$), industrial chemicals ($p=0.030$), and marginally significant associations with wine consumption ($p=0.056$) and degreasing chemicals ($p=0.091$). The mean value of alkaline phosphatase depended significantly on all four of these covariates.

The CC-adjusted analysis also showed a significantly higher mean value of alkaline phosphatase in the Ranch Hand group ($p=0.008$). The model was adjusted by the significant covariates of wine consumption (WINE) ($p<0.001$), occupation ($p<0.001$), and the interactions of age-by-industrial chemicals ($p=0.010$) and race-by-industrial chemicals ($p=0.007$). Wine consumption was used as a covariate instead of alcohol intensity since wine showed a very strong negative association with alkaline phosphatase. This effect masked a very weak positive association between beer or liquor consumption and alkaline phosphatase.

In the CD model a significant group-by-industrial chemicals interaction was found ($p=0.011$). Specifically, in those individuals exposed to industrial chemicals, the Ranch Hands had a significantly higher mean value than the Comparisons ($p<0.001$), whereas in the unexposed stratum, the mean values were not significantly different between groups ($p=0.973$; see Table K-1 of Appendix K). The CD analysis was also adjusted by wine consumption ($p<0.001$), occupation ($p<0.001$), and the interactions of age-by-industrial chemicals ($p=0.019$) and race-by-industrial chemicals ($p=0.002$).

The DD model revealed a marginally significant group difference (Adj. RR: 1.44, 95% C.I.: [0.97,2.13], p=0.070) following adjustment by four significant interactions of wine-by-degreasing chemicals (p=0.006), age-by-industrial chemicals (p=0.005), race-by-industrial chemicals (p=0.004), and occupation-by-industrial chemicals (p=0.016).

Total Bilirubin

Both the continuous and categorical unadjusted analyses found no significant differences in total bilirubin values between groups (p=0.576 and p=0.706, respectively).

The covariate associations for both groups showed a significant effect of race (8.5% abnormal in Blacks versus 2.5% in nonblacks; p<0.001). Significant differences in mean total bilirubin levels were found between occupational groups (0.76 mg/dl for officers, 0.72 mg/dl for enlisted flyers, and 0.75 mg/dl for enlisted groundcrew; p=0.011), and with increasing levels of current alcohol use (0.80 for more than four drinks per day, 0.75 for more than one to four drinks per day, and 0.74 for one or less drinks per day; p=0.008). Further, increasing levels of total bilirubin were marginally associated with age (p=0.093).

The CC model, adjusted for the interactions of age-by-degreasing chemicals (p=0.039), race-by-current alcohol use (p=0.007), and race-by-occupation (p=0.001), revealed no significant differences in total bilirubin means between groups (p=0.599). Similarly, the CD analysis found no difference between group means (p=0.598) after adjustment for the interactions of race-by-current alcohol use (p=0.004), occupation-by-current alcohol use (p=0.034), and occupation by race (p=0.002). The DD model, adjusted for race (p<0.001), also failed to detect significant group differences in the proportion of total bilirubin abnormalities (p=0.648).

Direct Bilirubin

Neither the continuous nor the categorical unadjusted tests disclosed significant differences between the Ranch Hand and Comparison groups (p=0.981 and p=0.649, respectively).

A covariate association with the categorical data combined from both groups was noted for race, with 7.8 percent abnormalities found in Blacks as contrasted to 3.3 percent in nonblacks (p=0.015). There was a significant association between mean values of direct bilirubin and current alcohol use (0.21 mg/dl, 0.17 mg/dl, and 0.17 mg/dl for more than four drinks per day, more than one to four drinks per day, and one or less drinks per day, respectively; p<0.001) and a marginally significant difference due to race (0.20 mg/dl for Blacks versus 0.18 mg/dl for nonblacks; p=0.059).

For both the CC and CD analyses, no significant group differences were found (p=0.972 and p=0.830, respectively). The CC model was adjusted for a race-by-current alcohol use interaction (p=0.025), and the CD model was adjusted for the significant interactions of race-by-current alcohol use (p=0.019), occupation-by-current alcohol use (p=0.002), current alcohol use-by-degreasing chemicals (p=0.012), and degreasing chemicals-by-industrial chemicals (p=0.025). The DD analysis revealed a group-by-industrial chemical

exposure interaction (p=0.012). For participants exposed to industrial chemicals, the Ranch Hands had a higher proportion with abnormal values than the Comparisons (5.3% abnormal versus 2.9%, respectively; p=0.035), whereas there was no group difference for participants not exposed to industrial chemicals (p=0.144). Each stratum of the interaction was adjusted for race (p=0.014) and current alcohol use (p=0.026). The biological relevance of this interaction is unclear at this time.

Lactic Dehydrogenase (LDH)

No significant differences were found between the groups, either in the proportion of abnormal values (p=0.560) or in the mean levels of LDH (p=0.655). Significant effects for age (121.6 U/L, 124.6 U/L, 135.3 U/L for those born in or after 1942, between 1923 and 1941, and in or before 1922, respectively; p<0.001) and race (129.5 U/L for Blacks versus 123.4 U/L for nonblacks; p=0.006) were found in the tests of mean LDH levels.

The CC analysis revealed a group-by-age interaction (p=0.018), although no significant adjusted group differences were found for any of the three age strata. The model was also adjusted for the significant interactions of occupation-by-exposure to industrial chemicals (p=0.014) and race by exposure to industrial chemicals (p=0.024). The CD model revealed no significant group differences after adjustment by age (p<0.001), race (p<0.001), and degreasing chemicals (p=0.016). Similarly, the DD analysis found no significant group differences, and no covariates made a significant contribution to the model.

Cholesterol

No significant differences were found between groups, either in the proportion of abnormal cholesterol levels (p=0.322) or in mean values of cholesterol (p=0.688) by unadjusted tests. However, in contrast, analysis of the Ranch Hand group versus the Original Comparisons (see Table K-9 of Appendix K) showed that the Comparisons had a significantly higher proportion of abnormal levels than the Ranch Hands (18.3% versus 14.5%, respectively; Est. RR: 0.76, 95% C.I.: [0.60,0.96], p=0.023). This observation was also found at Baseline. Significant covariate associations were noted between the proportion of participants with abnormally high cholesterol levels and age (12.7% for those born in or after 1942, 17.2% for those born between 1923 and 1941, and 18.4% for those born in or before 1922; p=0.010), occupation (14.9% for officers, 20.5% for enlisted flyers, and 13.9% for enlisted groundcrew; p=0.008), and current alcohol use (14.1% for one or less drinks per day, 16.4% for more than one to four drinks per day, and 21.7% for more than four drinks per day; p=0.018). For the associations between mean cholesterol levels and age, occupation, and current alcohol use, the significance of the covariate effects was greater than for the discrete analyses (p<0.001, p=0.002, and p<0.001, respectively).

The CC results showed no significant group difference (p=0.604). The model was adjusted by age (p<0.001), current alcohol use (p<0.001), industrial chemical exposure (p=0.043), and the race-by-degreasing chemicals (p=0.021) and race-by-occupation (p=0.005) interactions. The CD analysis was negative for significant group differences (p=0.548). The analysis included the covariate contributions made by age (p<0.001), current alcohol use

($p<0.001$), and a race-by-occupation interaction ($p=0.027$). The DD analysis also showed no significant difference between groups for adjusted proportions of participants with abnormal cholesterol levels ($p=0.181$). Contributing covariates included age ($p=0.029$), occupation ($p=0.039$), and a race-by-current alcohol use interaction ($p=0.012$). In all of the discrete cholesterol analyses, the cutpoint of 260 mg/dl was used.

Triglycerides

In the unadjusted analyses, no significant differences in the proportion of participants with abnormal triglyceride levels or in mean values were found between the Ranch Hand and Comparison groups ($p=0.549$ and $p=0.719$, respectively).

The covariate tests of association for percent abnormal triglycerides disclosed the significant effect of race (2.1% for Blacks and 6.7% for nonblacks; $p=0.031$) and a marginally significant association for industrial chemical exposure ($p=0.073$). For mean triglyceride levels, significant associations for age ($p<0.001$), race ($p<0.001$), occupation ($p=0.013$), current alcohol use ($p=0.030$), and degreasing chemicals ($p=0.019$) were noted, in addition to a marginally significant association with exposure to industrial chemicals ($p=0.077$).

The CC analysis revealed a significant group-by-age interaction ($p=0.015$), which showed a significantly elevated mean triglyceride level in Ranch Hands ($p=0.039$) born in or before 1922 as compared to similarly aged Comparisons (see Table K-1 of Appendix K). There were no significant differences for the other two age strata. A significant adjusting covariate was occupation ($p<0.001$); in addition, the current alcohol use-by-degreasing chemicals ($p=0.005$) and race-by-current alcohol use ($p=0.031$) interactions were used for adjustment. The CD-adjusted analysis found no significant group differences ($p=0.905$). The model was adjusted by age ($p<0.001$), race ($p<0.001$), occupation ($p<0.001$), and current alcohol use ($p=0.038$).

The DD analysis found a significant group-by-occupation interaction ($p=0.027$). Stratification by occupation revealed a significant increase in the proportion of abnormal triglyceride levels for Ranch Hand officers (Adj. RR: 1.77, 95% C.I.: [1.04,3.01], $p=0.035$) but no significant group differences were discerned for the enlisted flyer or enlisted groundcrew strata. The models were adjusted by race ($p=0.026$) and industrial chemical exposure ($p=0.038$). A cutpoint of 320 mg/dl was used to distinguish abnormal from normal.

Uroporphyrin

The uroporphyrin variable was analyzed only in the continuous form. The unadjusted analysis revealed a significant difference between group means (Comparisons 17.9 mg/24 hrs, Ranch Hands 16.9 mg/24 hrs; $p=0.048$).

A CC model found a significant group-by-blood urea nitrogen (BUN) interaction ($p=0.015$; see Table K-1 of Appendix K). To interpret the interaction, BUN was dichotomized at the median value of 14 mg/dl. Stratifying by BUN levels revealed a significantly greater ($p<0.001$) uroporphyrin mean for Comparisons than for Ranch Hands for BUN levels of 14 or less mg/dl and a non-significant but greater Ranch Hand mean for participants with BUN levels of

more than 14 mg/dl. The stratified model was adjusted for current alcohol use (p=0.026) and the occupation-by-degreasing chemicals (p=0.005) interaction.

Coproporphyrin

As with the uroporphyrin variable, coproporphyrin was analyzed only as a continuous variable. The unadjusted analysis revealed a borderline significant difference in the mean coproporphyrin levels (119.1 mg/24 hrs for Ranch Hands and 115.6 mg/24 hrs for Comparisons; p=0.081).

The covariate tests of association detected the significant effects of age (p=0.003) and current alcohol use (p=<0.001).

A CC model, adjusted by BUN (p<0.001) and an age-by-current alcohol use interaction (p=0.003) revealed a borderline significant group difference (p=0.065) similar to the unadjusted analysis. The adjusted coproporphyrin means were 119.3 mg/24 hrs and 115.7 mg/24 hrs for the Ranch Hands and Comparisons, respectively.

Discussion

The results from the nine hepatic and two porphyrin analyses were not totally consistent with the Baseline findings. Several analytical reasons may possibly explain some of these differences, i.e., the adjusted analyses herein used the additional covariates of age, race, and occupation (the matching variables), and all two-way covariate interactions. However, as the Baseline data were not reanalyzed with the model process and total Comparison group used in this report, the contribution of analytic technique versus a true change in hepatic status is unknown.

The Baseline Report noted a significantly lower mean cholesterol level in the Ranch Hands (opposite of an expected dioxin effect) and slight tendencies for higher GGTP and LDH values in the Ranch Hands. In this chapter, the analyses have shown equivalent group cholesterol levels, an increased SGPT mean in the Comparisons, an increased mean alkaline phosphatase in the Ranch Hands, an increased uroporphyrin mean in the Comparisons, and a borderline increased coproporphyrin mean in the Ranch Hands. The individual hepatic assay results were not suggestive of a pattern of significant hepatic damage in the Ranch Hands that might be supportive of an herbicide effect. Further, there was no consistent group-by-covariate interaction that suggests a detriment to a specific subcategory of the Ranch Hands.

For those covariates used in both the Baseline study and this followup study, the direction and magnitude of their effects were relatively consistent between the studies. However, an unexpected association between wine drinking and alkaline phosphatase lacks a plausible explanation, particularly considering the inverse relationship, i.e., increasing alkaline phosphatase levels with decreasing wine consumption. These findings suggested the association between wine and alkaline phosphatase may be due to imprecision in data collection.

None of the categorical (normal/abnormal categories) analyses was statistically significant, whereas all of the significant results were

generated by the more powerful contrasts of continuously distributed hepatic data.

Both porphyrin analyses showed group associations and are in distinct contrast to the otherwise largely negative hepatic findings. The significantly elevated uroporphyrin mean value in the Comparisons was directly opposite to that expected if dioxin-induced PCT were prevalent in the Ranch Hands. The primary biochemical defect in PCT is the reduced activity of uroporphyrinogen decarboxylase, an enzyme that metabolizes uroporphyrin. This defect leads to increased levels of uroporphyrin and coproporphyrin.

Questionnaire-Laboratory Correlations: Porphyria Cutanea Tarda

In the interval questionnaire all participants were asked whether their skin manifested "patches," excessive bruises, or sensitivity. These questions were deemed important in order to bound the maximum prevalence of cutaneous disorders compatible with a diagnosis of PCT. These historical data are given in Table 13-13.

TABLE 13-13.

Unadjusted Analysis for Interval History of Skin Bruises, Skin Patches, and Skin Sensitivity by Group

	Bruises, Patches, or Sensitivity					
	Yes		No			
Group	Number	Percent	Number	Percent	Total	p-Value
Ranch Hand	265	26.2	746	73.8	1,011	0.001
Comparison	260	20.2	1,029	79.8	1,289	

These data revealed that the Ranch Hands reported significantly more cutaneous symptoms (26.2%) than the Comparisons (20.2%). However, these data were substantially less than those reported at the Baseline in-home questionnaire, which also showed a statistically significant excess in the Ranch Hands.

To determine if the skin histories might be related to PCT, the historic data were compared to the porphyrin test results. The abnormal/normal cut-point of the coproporphyrin assays was reset to the 95th percentile because the normal range of the laboratory overclassified the proportion of abnormals. Table 13-14 gives the tabular display of both porphyrin test results by the reporting history of skin disorders.

TABLE 13-14.

Unadjusted Analyses for Porphyrin Abnormalities by Group and Skin Patch, Bruise, or Sensitivity Reported at Followup Questionnaire

Group	Skin Patch, Bruise, or Sensitivity Reported	Abnormal Porphyrin Findings for a Participant: 0 Number	0 Percent	1 Number	1 Percent	2 Number	2 Percent	Total	p-Value*
Both Groups	Yes	472	90.1	48	9.2	4	0.8	524	0.789
	No	1,593	90.2	165	9.3	9	0.5	1,767	
Ranch Hand	Yes	239	90.5	24	9.1	1	0.4	264	0.950
	No	670	90.3	70	9.4	2	0.3	742	
Comparison	Yes	233	89.6	24	9.2	3	1.2	260	0.742
	No	923	90.1	95	9.3	7	0.7	1,025	

*Chi-square test, 2 d.f.

The data from both groups combined suggest that there is no relationship between a history of cutaneous disorders and porphyrin test positivity. The group-specific data in the table also show a lack of a statistically significant association between the reporting of skin patches, bruises, or sensitivity and the presence of an abnormal porphyrin test result. However, in both the Ranch Hand and Comparison groups, participants who had abnormal tests for both uroporphyrins and coproporphyrins were more likely to have reported cutaneous disorders than participants with normal findings for both tests. Consequently, the data were retabulated, focusing only upon uroporphyrin abnormalities (absolutely required for a diagnosis of PCT) and reporting of cutaneous disorders. These data are summarized in Table 13-15.

These data suggest that the relative risk of increased uroporphyrin abnormalities for Ranch Hands is independent of whether or not a study participant reported skin patches, bruises, or sensitivities at the followup questionnaire (Breslow-Day test of homogeneity of odds ratio, p=0.791). In each instance (reported/not reported), the estimated relative risk was nonsignificant and less than 1, and in both the Ranch Hand group and the Comparison group there was a higher percentage of uroporphyrin abnormalities for participants who did not report skin patches, bruises, or sensitivity than for participants who did report these conditions.

Thus, the sequential displays of Tables 13-13 through 13-15 show excessive reporting of PCT-like cutaneous symptoms in the Ranch Hand group that was not related to test abnormalities for both uroporphyrin and coproporphyrin abnormal test results, or for uroporphyrin abnormalities alone. These analyses were consistent with the clinical observation that

TABLE 13-15.

Unadjusted Analyses for Uroporphyrin Abnormalities by Group and Skin Patch, Bruise, or Sensitivity Reported at Followup Questionnaire

Variable	Stratification	Statistic	Group: Ranch Hand Number	Ranch Hand Percent	Group: Comparison Number	Comparison Percent	Est. Relative Risk (95% C.I.)	p-Value
Uroporphyrin								
Skin Patch, Bruise, or Sensitivity Reported		n	264		260			
		Abnormal	12	4.5	12	4.6	0.98 (0.43,2.23)	0.999
		Normal	252	95.5	248	95.4		
Skin Patch, Bruise, or Sensitivity Not Reported		n	742		1,025			
		Abnormal	42	5.7	66	6.4	0.89 (0.62,1.28)	0.547
		Normal	700	94.3	959	93.6		

only one differential diagnosis at the examination entertained the diagnosis of PCT. Based on all of these observations, PCT was a rare, if not non-existent, condition in the Ranch Hands.

EXPOSURE INDEX ANALYSES

Both unadjusted and adjusted exposure index analyses were carried out for the nine laboratory tests of hepatic function and the two porphyrin metabolite tests. The porphyrin variables were analyzed only as continuous variables, while the others were analyzed both as continuous variables and discretized variables. Five covariates were included in the adjusted analyses: age, race, current alcohol use, exposure to degreasing chemicals (yes/no), and exposure to industrial chemicals (yes/no). Current alcohol use was treated as a continuous variable for all adjusted analyses, and age was treated as a continuous variable for the continuous adjusted analyses. Age was trichotomized (born in or after 1942, born between 1923 and 1941, and born in or before 1922) for the discrete adjusted analyses. In addition, the covariate BUN was used in the porphyrin analyses.

For each variable, exposure level frequencies and percents are presented in Table K-3 of Appendix K along with the results of the unadjusted discrete

analyses using Pearson's chi-square test to reflect overall exposure index differences and Fisher's exact test to investigate medium versus low and high versus low exposure level contrasts. Unadjusted means for each exposure level are presented in Table K-4 of Appendix K, along with the results of the unadjusted continuous analyses (using an F-test for an overall group assessment) and t-tests to examine medium versus low and high versus low exposure index contrasts. Results of the adjusted categorical and adjusted continuous analyses are presented in Tables 13-16 and 13-17, respectively. These results are presented in the context of a main effects model containing exposure index and all five covariates. Additional adjusted continuous analyses were conducted to examine pairwise interactions involving the exposure index and the covariates. Unadjusted and adjusted results for each variable are discussed in sequence.

SGOT

Within each occupation cohort, the low exposure level had the lowest percentage of abnormalities and the lowest mean. A marginally significant overall exposure level relationship (p=0.065) was found in the unadjusted discrete analysis for the enlisted groundcrew. This association was statistically significant in the adjusted analysis (p=0.023), exhibiting a dose-response effect; medium versus low (Adj. RR: 2.14, 95% C.I.: [0.77,5.99], p=0.147) and high versus low (Adj. RR: 3.64, 95% C.I.: [1.36,9.72], p=0.010). A nonsignificant dose-response relationship was observed in the corresponding unadjusted and adjusted continuous analyses (p=0.418 and p=0.409, respectively), with unadjusted means of 32.9 U/L, 33.2 U/L, and 34.4 U/L for the low, medium, and high exposure levels, respectively. No significant results were found for enlisted flyers and officers.

SGPT

Within the enlisted groundcrew and enlisted flyer cohorts the low exposure level had the lowest percentage of abnormalities and the lowest mean value. This situation was reversed for the officers who exhibited the highest percentage of abnormal measurements and highest mean value in the low exposure categories.[1] A significant overall result was found for enlisted flyers in the adjusted discrete analysis (p=0.036; medium versus low, Adj. RR: 6.55, 95% C.I.: [1.25,34.43], p=0.026); high versus low, Adj. RR: 4.29, 95% C.I.: [0.75,24.35], p=0.101). In the corresponding adjusted continuous analyses, a marginally significant dose-response relationship was observed (p=0.058) with adjusted means 18.1 U/L, 21.4 U/L, and 21.8 U/L for the low, medium, and high exposure levels, respectively. No significant results were found for officers or enlisted groundcrew.

GGTP

No significant or marginally significant results were found. A non-significant dose-response relationship was seen for enlisted flyers and officers in the continuous analyses but, conversely, a nonsignificant decreasing dose-response relationship was seen in the enlisted groundcrew.

TABLE 13-16.

Adjusted Categorical Exposure Index Analyses (Main Effects Model) Results for Hepatic Function Variables by Occupation

Variable	Occupation	Exposure Index: Low Total	Exposure Index: Medium Total	Exposure Index: High Total	Contrast	Adj. Relative Risk (95% C.I.)	p-Value
SGOT	Officer	125	129	120	Overall		0.508
					M vs. L	1.60 (0.64,3.98)	0.312
					H vs. L	1.02 (0.38,2.77)	0.963
	Enlisted Flyer	55	65	57	Overall		0.108
					M vs. L	7.79 (0.77,79.20)	0.083
					H vs. L	5.38 (0.49,59.50)	0.170
	Enlisted Groundcrew	152	160	140	Overall		0.023
					M vs. L	2.14 (0.77,5.99)	0.147
					H vs. L	3.64 (1.36,9.72)	0.010
SGPT	Officer	125	129	120	Overall		0.768
					M vs. L	0.97 (0.48,1.97)	0.933
					H vs. L	0.77 (0.37,1.64)	0.504
	Flyer	Enlisted	55	65	57	Overall	0.036
					M vs. L	6.55 (1.25,34.43)	0.026
					H vs. L	4.29 (0.75,24.35)	0.101
	Enlisted Groundcrew	152	160	140	Overall		0.457
					M vs. L	1.53 (0.77,3.01)	0.223
					H vs. L	1.18 (0.57,2.48)	0.655

TABLE 13-16. (continued)

Adjusted Categorical Exposure Index Analyses (Main Effects Model) Results for Hepatic Function Variables by Occupation

Variable	Occupation	Exposure Index: Low Total	Exposure Index: Medium Total	Exposure Index: High Total	Contrast	Adj. Relative Risk (95% C.I.)	p-Value
	Officer	125	129	120	Overall		0.987
					M vs. L	1.02 (0.38,2.72)	0.968
					H vs. L	0.94 (0.35,2.54)	0.906
GGTP	Enlisted Flyer	55	65	57	Overall		0.798
					M vs. L	1.51 (0.41,5.65)	0.536
					H vs. L	1.46 (0.37,5.78)	0.586
	Enlisted Groundcrew	152	160	140	Overall		0.760
					M vs. L	0.74 (0.34,1.64)	0.462
					H vs. L	0.89 (0.40,1.97)	0.776
	Officer	126	129	120	Overall		0.272
					M vs. L	2.44 (0.65,9.05)	0.184
					H vs. L	0.91 (0.19,4.36)	0.926
Alkaline Phosphatase	Enlisted Flyer	54	64	56	Overall		0.191
					M vs. L	4.84 (0.52,44.80)	0.165
					H vs. L	5.34 (0.58,49.06)	0.139
	Enlisted Groundcrew	153	160	141	Overall		0.431
					M vs. L	1.35 (0.50,3.59)	0.552
					H vs. L	1.82 (0.72,4.59)	0.202

TABLE 13-16. (continued)

Adjusted Categorical Exposure Index Analyses (Main Effects Model) Results for Hepatic Function Variables by Occupation

Variable	Occupation	Exposure Index: Low Total	Exposure Index: Medium Total	Exposure Index: High Total	Contrast	Adj. Relative Risk (95% C.I.)	p-Value
Total Bilirubin	Officer	125	129	120	Overall		0.851
					M vs. L	0.67 (0.10,4.51)	0.677
					H vs. L	1.10 (0.18,6.61)	0.915
	Enlisted Flyer[a]	54	65	57	--	--	--
	Enlisted Groundcrew	152	160	140	Overall		0.332
					M vs. L	0.41 (0.10,1.65)	0.208
					H vs. L	1.02 (0.32,3.23)	0.971
Direct Bilirubin	Officer	125	129	120	Overall		0.354
					M vs. L	2.69 (0.46,15.82)	0.274
					H vs. L	3.10 (0.56,17.25)	0.196
	Enlisted Flyer	55	65	57	Overall		0.466
					M vs. L	2.97 (0.48,18.38)	0.241
					H vs. L	1.79 (0.24,13.43)	0.571
	Enlisted Groundcrew	152	160	140	Overall		0.767
					M vs. L	1.61 (0.43,6.06)	0.480
					H vs. L	1.40 (0.36,5.51)	0.628

TABLE 13-16. (continued)

Adjusted Categorical Exposure Index Analyses (Main Effects Model) Results for Hepatic Function Variables by Occupation

Variable	Occupation	Exposure Index: Low Total	Exposure Index: Medium Total	Exposure Index: High Total	Contrast	Adj. Relative Risk (95% C.I.)	p-Value
	Officer	125	129	120	Overall		0.107
					M vs. L	0.54 (0.27,1.09)	0.085
					H vs. L	0.50 (0.25,1.03)	0.060
Cholesterol	Enlisted Flyer	55	65	57	Overall		0.972
					M vs. L	1.02 (0.38,2.73)	0.962
					H vs. L	1.12 (0.42,3.02)	0.822
	Enlisted Groundcrew	152	160	140	Overall		0.417
					M vs. L	1.20 (0.57,2.55)	0.630
					H vs. L	1.61 (0.78,3.30)	0.194
	Officer	125	129	120	Overall		0.721
					M vs. L	0.97 (0.38,2.45)	0.946
					H vs. L	1.35 (0.55,3.32)	0.514
Triglycerides	Enlisted Flyer	55	65	57	Overall		0.379
					M vs. L	2.66 (0.62,11.39)	0.189
					H vs. L	2.06 (0.44,9.60)	0.358
	Enlisted Groundcrew	152	160	140	Overall		0.363
					M vs. L	0.44 (0.14,1.42)	0.173
					H vs. L	0.60 (0.19,1.86)	0.375

[a]No analysis done since there were only three abnormal (one medium, two high).

TABLE 13-17.

Adjusted Continuous Exposure Index Analyses (Main Effects Model) for Hepatic Function Variables and Two Porphyrin Determinations by Occupation

Variable	Occupation	Statistic	Exposure Index Low	Medium	High	Contrast	p-Value
SGOT	Officer	n	125	129	120	Overall	0.718
		Adj. Mean	33.6	34.7	33.8	M vs. L	0.450
						H vs. L	0.904
	Enlisted Flyer	n	55	65	57	Overall	0.276
		Adj. Mean	30.3	32.8	32.7	M vs. L	0.144
						H vs. L	0.184
	Enlisted Groundcrew	n	152	160	140	Overall	0.409
		Adj. Mean	33.5	34.1	35.0	M vs. L	0.595
						H vs. L	0.183
SGPT	Officer	n	125	129	120	Overall	0.695
		Adj. Mean	20.1	20.0	19.1	M vs. L	0.969
						H vs. L	0.451
	Enlisted Flyer	n	55	65	57	Overall	0.058
		Adj. Mean	18.1	21.4	21.8	M vs. L	0.047
						H vs. L	0.030
	Enlisted Groundcrew	n	152	160	140	Overall	0.581
		Adj. Mean	20.2	21.4	21.0	M vs. L	0.309
						H vs. L	0.492

TABLE 13-17. (continued)

Adjusted Continuous Exposure Index Analyses (Main Effects Model) for Hepatic Function Variables and Two Porphyrin Determinations by Occupation

Variable	Occupation	Statistic	Exposure Index Low	Medium	High	Contrast	p-Value
GGTP	Officer	n	125	129	120	Overall	0.828
		Adj. Mean	30.9	32.2	32.4	M vs. L	0.611
						H vs. L	0.580
	Enlisted Flyer	n	55	65	57	Overall	0.286
		Adj. Mean	36.6	42.6	44.6	M vs. L	0.230
						H vs. L	0.132
	Enlisted Groundcrew	n	152	160	140	Overall	0.299
		Adj. Mean	36.9	36.6	33.1	M vs. L	0.914
						H vs. L	0.159
Alkaline Phosphatase	Officer	n	126	129	120	Overall	0.843
		Adj. Mean	82.3	82.9	83.8	M vs. L	0.808
						H vs. L	0.561
	Enlisted Flyer	n	54	64	56	Overall	0.127
		Adj. Mean	90.7	99.3	97.7	M vs. L	0.053
						H vs. L	0.122
	Enlisted Groundcrew	n	153	160	141	Overall	0.576
		Adj. Mean	91.5	94.0	93.5	M vs. L	0.318
						H vs. L	0.444

TABLE 13-17. (continued)

Adjusted Continuous Exposure Index Analyses (Main Effects Model) for Hepatic Function Variables and Two Porphyrin Determinations by Occupation

Variable	Occupation	Statistic	Exposure Index Low	Medium	High	Contrast	p-Value
Total Bilirubin	Officer	n	125	129	120	Overall	0.439
		Adj. Mean	0.77	0.75	0.79	M vs. L	0.504
						H vs. L	0.545
	Enlisted Flyer	n	55	65	57	Overall	0.070
		Adj. Mean	0.69	0.76	0.79	M vs. L	0.128
						H vs. L	0.023
	Enlisted Groundcrew	n	152	160	140	Overall	0.240
		Adj. Mean	0.73	0.74	0.78	M vs. L	0.838
						H vs. L	0.117
Direct Bilirubin	Officer	n	125	129	120	Overall	0.567
		Adj. Mean	0.20	0.19	0.21	M vs. L	0.517
						H vs. L	0.689
	Enlisted Flyer	n	55	65	57	Overall	0.724
		Adj. Mean	0.18	0.19	0.19	M vs. L	0.471
						H vs. L	0.498
	Enlisted Groundcrew	n	152	160	140	Overall	0.550
		Adj. Mean	0.17	0.19	0.18	M vs. L	0.277
						H vs. L	0.670

TABLE 13-17. (continued)

Adjusted Continuous Exposure Index Analyses (Main Effects Model) for Hepatic Function Variables and Two Porphyrin Determinations by Occupation

Variable	Occupation	Statistic	Exposure Index Low	Medium	High	Contrast	p-Value
LDH	Officer	n	125	129	120	Overall	0.232
		Adj. Mean	134.0	131.3	128.9	M vs. L	0.373
						H vs. L	0.088
	Enlisted Flyer	n	55	65	57	Overall	0.101
		Adj. Mean	114.4	112.4	120.9	M vs. L	0.619
						H vs. L	0.129
	Enlisted Groundcrew	n	152	160	140	Overall	0.092
		Adj. Mean	125.3	125.3	129.7	M vs. L	0.997
						H vs. L	0.055
Cholesterol	Officer	n	125	129	120	Overall	0.049
		Adj. Mean	236.7	225.0	224.2	M vs. L	0.039
						H vs. L	0.029
	Enlisted Flyer	n	55	65	57	Overall	0.214
		Adj. Mean	213.2	208.1	220.6	M vs. L	0.492
						H vs. L	0.343
	Enlisted Groundcrew	n	152	160	140	Overall	0.945
		Adj. Mean	209.6	211.1	210.1	M vs. L	0.742
						H vs. L	0.927

TABLE 13-17. (continued)

Adjusted Continuous Exposure Index Analyses (Main Effects Model) for Hepatic Function Variables and Two Porphyrin Determinations by Occupation

Variable	Occupation	Statistic	Exposure Index Low	Medium	High	Contrast	p-Value
Triglycerides	Officer	n	125	129	120	Overall	0.739
		Adj. Mean	110.0	108.5	116.3	M vs. L	0.886
						H vs. L	0.558
	Enlisted Flyer	n	55	65	57	Overall	0.981
		Adj. Mean	112.5	111.2	113.7	M vs. L	0.919
						H vs. L	0.927
	Enlisted Groundcrew	n	152	160	140	Overall	0.922
		Adj. Mean	110.9	109.9	107.9	M vs. L	0.890
						H vs. L	0.690
Uroporphyrin	Officer	n	125	129	120	Overall	0.856
		Adj. Mean	17.49	16.69	17.45	M vs. L	0.621
						H vs. L	0.977
	Enlisted Flyer	n	54	65	57	Overall	0.703
		Adj. Mean	18.58	16.96	18.27	M vs. L	0.438
						H vs. L	0.890
	Enlisted Groundcrew	n	151	160	139	Overall	0.644
		Adj. Mean	16.39	16.54	15.45	M vs. L	0.903
						H vs. L	0.451

TABLE 13-17. (continued)

Adjusted Continuous Exposure Index Analyses (Main Effects Model) for Hepatic Function Variables and Two Porphyrin Determinations by Occupation

Variable	Occupation	Statistic	Exposure Index Low	Medium	High	Contrast	p-Value
	Officer	n	125	129	120	Overall	0.901
		Adj. Mean	127.65	128.84	130.26	M vs. L	0.833
						H vs. L	0.649
Coproporphyrin	Enlisted	n	55	65	57	Overall	0.669
	Flyer	Adj. Mean	108.67	115.31	109.81	M vs. L	0.408
						H vs. L	0.890
	Enlisted	n	151	160	140	Overall	0.325
	Groundcrew	Adj. Mean	115.28	115.71	122.88	M vs. L	0.935
						H vs. L	0.177

Alkaline Phosphatase

For the enlisted groundcrew and the enlisted flyers, the lowest abnormal prevalence rate and lowest mean value were found in the low exposure category. A nonsignificant increasing dose-response relationship was seen within these occupations for the discrete analyses. In both unadjusted and adjusted continuous analyses, a marginally significant medium versus low contrast was found for enlisted flyers ($p=0.086$ and $p=0.053$, respectively), with unadjusted means of 88.9 U/L, 96.3 U/L, and 95.2 U/L for the low, medium, and high exposure levels, respectively.

Total Bilirubin

Discrete analyses revealed no significant findings; adjusted discrete analyses for enlisted flyers were not done due to sparse data. Continuous analyses revealed a significant overall effect ($p=0.045$, unadjusted) for enlisted flyers, which was marginally significant after adjustment ($p=0.070$). In both unadjusted and adjusted analyses, the high versus low mean contrast was significant ($p=0.014$ and $p=0.023$, respectively), with unadjusted means of 0.66 mg/dl, 0.73 mg/dl, and 0.76 mg/dl for the low, medium, and high exposure levels, respectively.

Direct Bilirubin

There were no significant exposure findings in either the continuous or discrete analyses, although within each occupational cohort, the lowest abnormal prevalence rate was found in the low exposure group.

LDH

The unadjusted discrete analyses revealed no significant or marginally significant results. No adjusted discrete analyses were done due to sparse data. The unadjusted continuous analyses for the enlisted groundcrew showed a significant overall relationship with the exposure index ($p=0.031$), with mean values of 123.1 U/L, 122.3 U/L, 127.9 U/L for the low, medium, and high exposure levels; the high versus low contrast was significant ($p=0.037$). After adjustment, the continuous analyses for enlisted groundcrew revealed marginally significant results ($p=0.092$, overall; $p=0.055$, high versus low). No significant or marginally significant results were seen for enlisted flyers or officers. Enlisted flyers and enlisted groundcrew had the largest mean values for their highest exposure category, which is reversed in the officers, who exhibited a nonsignificant decreasing dose-response relationship with exposure level.

Cholesterol

Significant or marginally significant results were found for officers in the direction of a decreasing dose-response relationship in both the adjusted continuous (overall $p=0.049$, medium versus low $p=0.039$, high versus low $p=0.029$) and adjusted discrete (medium versus low $p=0.085$, high versus low $p=0.060$) analyses. Neither of the enlisted cohorts demonstrated a similar decreasing response.

Triglycerides

No significant or marginally significant results were found.

Uroporphyrins and Coproporphyrins

No significant or marginally significant results were found.

EXPOSURE INDEX ANALYSES

Additional continuous analyses were done to examine pairwise interactions involving exposure level and the covariates. Ten exposure group-by-covariate interactions were found at $p \leq 0.05$. All interactions were found in the enlisted flyer and enlisted groundcrew occupations. Eight of the interactions involved current alcohol consumption, one involved age, and one involved race. The interactions are summarized in Tables K-5 and K-6 of Appendix K. In Table K-5 of Appendix K, the slope of the continuous covariate with respect to the dependent variable is provided for each of the three exposure levels. Table K-6 of Appendix K presents the mean level of direct bilirubin for each of the three exposure levels by race. The interactions involving current alcohol consumption are mainly due to a nonsignificant dependent variable response to increasing alcohol consumption in the low exposure group in contrast to a significant positive response for the medium and high groups. The SGOT, SGPT, and GGTP interaction results for the enlisted groundcrew provide support for an interpretation of herbicide effect.

In summary, the nine hepatic function variables and two porphyrin metabolite variables showed no conclusive evidence of a dose-response relationship at the followup examination. Five overall exposure group differences were found. Only two of these (SGOT for enlisted groundcrew, and total bilirubin for enlisted flyers) supported a dose-response relationship.

LONGITUDINAL ANALYSES

Three hepatic enzyme variables, SGOT, SGPT, and GGTP, were chosen for longitudinal analysis, spanning the spectrum of intermediate to acute effects. These test variables were chosen because both the Baseline and the followup assays were performed by the high-precision ACA 500® DuPont technology. The data from these three hepatic variables are arrayed in Table 13-18.

The SGOT and SGPT data showed slight but uniform increases from the Baseline examination. These increases were proportionately the same for both the Ranch Hand and Comparison groups. These changes may reflect an aging effect or are due to laboratory variation. As indicated by the equality-of-difference p-values, none of the three hepatic variables showed a statistically significant difference in the changes from Baseline to followup between groups.

TABLE 13-18.

Longitudinal Analyses for SGOT, SGPT, and GGTP:
A Contrast of Baseline and First Followup Examination Test Means

Variable	Group	Total	Means 1982 Baseline	Means 1985 Followup	p-Value* (Equality of Difference)
SGOT	Ranch Hand	971	32.91	33.73	0.61
	Comparison	1,139	32.97	33.73	
SGPT	Ranch Hand	971	20.08	21.82	0.72
	Comparison	1,139	20.51	22.44	
GGTP	Ranch Hand	971	39.26	33.16	0.63
	Comparison	1,139	38.64	32.35	

*Analyzed in log units.

SUMMARY AND CONCLUSIONS

The interval questionnaire revealed sparse reporting of liver disorders from 1982 to 1985 that was not significantly different between groups. Historical liver disease was verified by medical records, and these data were added to the verified Baseline history to assess possible lifetime differences. No significant differences were found. The medical record verification process showed that the historical data were generally correctly reported and classified between groups, except for the category of enlarged liver which showed a higher verification rate in the Comparison group.

Digestive system mortality showed an overall nonsignificant excess in the Ranch Hands, but a relative nonsignificant excess of malignant neoplasms in the Comparisons.

No differences were found for past or current peptic ulcer disease for the Ranch Hand and Comparison groups, adjusted for standard covariates as well as blood type.

The physical examination disclosed a borderline significant increase of hepatomegaly in the Ranch Hand group. Emphasis was placed on nine laboratory test variables measuring liver function, i.e., serum glutamic-oxaloacetic transaminase (SGOT), serum glutamic-pyruvic transaminase (SGPT), gamma-glutamyl transpeptidase (GGTP), alkaline phosphatase, total and direct bilirubin, lactic dehydrogenase (LDH), cholesterol, and triglycerides. In addition, uroporphyrin and coproporphyrin measurements were obtained to assess liver function and the likelihood of porphyria cutanea tarda (PCT). The nine hepatic variables were subjected to continuous and categorical statistical tests, and were adjusted for the covariates age, race, occupation, current alcohol consumption, and unprotected exposure to both industrial chemicals and degreasing chemicals. Final statistical models used only

the significant covariates and two-way interactions for adjustment. The two porphyrin measurements were analyzed only in the continuous form. The overall summary results of the analyses of these 11 variables are given in Table 13-19.

The results showed a significantly lower mean SGPT level, a greater mean alkaline phosphatase level, a lower mean uroporphyrin level for Ranch Hands as contrasted with Comparisons, and a marginally significant greater mean coproporphyrin level. Only in the instance of alkaline phosphatase did the discrete analysis approach statistical significance. No group differences were noted for SGOT, GGTP, total and direct bilirubin, LDH, cholesterol, or triglycerides. However, an analysis using only the Original Comparisons revealed a significantly greater mean cholesterol level in the Comparison group. A review of the covariate effects in the adjusted statistical models revealed that all covariates behaved as expected with the exception of alcohol consumption for the alkaline phosphatase analysis, which showed an inverse relationship with wine consumption.

Exploration of group-by-group covariate interactions for alkaline phosphatase, direct bilirubin, triglycerides, SGOT, and uroporphyrins revealed significant group differences within specific covariate strata. In particular, Ranch Hands exposed to industrial chemicals had a significantly higher adjusted mean level of alkaline phosphatase and a significantly higher abnormal prevalence rate of direct bilirubin than similarly exposed Comparisons. For triglycerides, Ranch Hands born in or before 1922 had a significantly higher adjusted mean level than similar aged Comparisons, while Ranch Hand officers exhibited a significantly higher abnormal prevalence rate than Comparison officers. For SGOT, Ranch Hand moderate current drinkers (more than one to four drinks per day) had a significantly higher mean level than corresponding Comparisons. In the opposite direction, Comparisons with a mean BUN level less than or equal to 14 (median for all participants) were found to have a significantly higher adjusted mean uroporphyrin level than similar Ranch Hands. These results did not disclose any common pattern detrimental to the Ranch Hand group.

These findings were generally consistent with the 1982 Baseline data, which disclosed a significantly increased mean cholesterol level in the Comparisons and nonsignificant Ranch Hand mean elevations for GGTP and LDH. Slight differences in analytic results are probably due to the use of more fully adjusted models used for the followup examination data.

Overall, the followup examination laboratory data showed no adverse clinical or exposure patterns in either group. Further, the detection of significant mean shifts (still within normal range) by the continuous statistical tests, not mirrored by the categorical tests, suggests a circumstance of statistical power rather than findings of biological relevance.

Of the five significant or marginally significant results that were found in the adjusted exposure index analyses, four exhibited a trend suggestive of an increasing dose-response relationship. In the enlisted flyer cohort, the percentages of SGPT abormalities were significantly different and increased from the low to the high exposure categories. The corresponding mean values were marginally significantly different among exposure levels. Also, the mean levels of total bilirubin were marginally significantly different among exposure levels, increasing with exposure level. For enlisted groundcrew, the percentage of SGOT abnormalities significantly differed among

TABLE 13-19.

Overall Summary Results of Unadjusted and Adjusted Analyses of Nine Hepatic Function Variables and Two Porphyrin Metabolite Tests

Variable	Unadjusted		Adjusted*			Direction of Results**
	Mean	Categorical	Mean CC	Mean CD	Categorical DD	
Questionnaire						
Liver Disease (Lifetime History)						
Hepatitis	--	NS	--	--	--	
Jaundice	--	NS	--	--	--	
Cirrhosis	--	NS	--	--	--	
Enlarged Liver	--	NS	--	--	--	
Miscellaneous Liver Disorders	--	NS	--	--	--	
Peptic Ulcer Disease	--	NS	--	--	NS[a]	
Physical Examination						
Hepatomegaly	--	NS*	--	--	--	RH>C
Laboratory Testing						
SGOT	NS	NS	NS	****	NS	
SGPT	NS*	NS	0.048	0.029	NS	C>RH
GGTP	NS	NS	NS	NS	NS	
Alkaline Phosphate	0.009	NS	0.008	****	NS*	RH>C
Total Bilirubin	NS	NS	NS	NS	NS	
Direct Bilirubin	NS	NS	NS	NS	****	
LDH	NS	NS	****	NS	NS	
Cholesterol	NS	NS	NS	NS	NS	
Triglycerides	NS	NS	****	NS	****	
Uroporphyrin	0.048	--	****	--	--	C>RH
Coproporphyrin	NS*	--	NS*	--	--	RH>C
Questionnaire-Laboratory Correlation						
Skin Bruises, Patches, and Sensitivity	--	0.001	--	--	--	RH>C

*C: Continuous
D: Discrete
**RH>C: more abnormalities, or higher mean value, in Ranch Hands.
C>RH: more abnormalities, or higher mean value, in Comparisons.
[a]Adjusted for blood type.
NS: Not significant ($p>0.10$).
NS*: Borderline significant ($0.05<p\leq0.10$).
--Analysis not performed.
****Group-by-covariate interaction.

exposure levels. Within the enlisted flyer cohort, all nine laboratory tests of hepatic function had the lowest percentage of abnormalities in the low exposure category; correspondingly, six of the nine mean levels were lowest for the low exposure category. Of the ten group-by-covariate interactions that were found, three (SGOT, SGPT, and GGTP) supported a dose-response relationship in the enlisted groundcrew cohort. Exploration of these interactions revealed a trend that showed an increasing association between current alcohol consumption and the dependent variables for increasing exposure levels.

Longitudinal analyses for SGOT, SGPT, and GGTP disclosed no statistically significant group differences in the mean shifts from the Baseline to the followup examination.

Interval reporting of PCT-like symptoms of skin patches, bruises, and sensitivity was significantly increased in the Ranch Hands ($p=0.001$). However, when these historic data were contrasted to both uroporphyrin and coproporphyrin abnormalities, no correlation was apparent, nor were there any significant group differences. Since an elevation in the uroporphyrin level is required for a diagnosis of PCT, the historic data were retabulated with only uroporphyrin abnormalities; again, no group differences were apparent, and, in fact, uroporphyrin abnormalities in both groups were higher in those participants without a history of skin disorders than in those participants with such a history. The likelihood of bona fide PCT among study participants, and particularly among the Ranch Hands, appears to be remote.

In conclusion, the followup examination disclosed more statistically significant findings for tests of liver function than the Baseline examination, but they were equally divided between the two groups and did not demonstrate clinical, statistical, or exposure patterns consistent with an herbicide-related effect on health. No evidence was found to suggest an increased likelihood of PCT among the Ranch Hand group.

CHAPTER 13

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